CLAIMS

What is claimed is:

1. A method for modifying a biomolecule, comprising:

a) immobilizing a biomolecule bound to a magnetic particle on a magnetic separation apparatus by applying a magnetic field to a magnetizable matrix in the column; and

b) modifying the immobilized biomolecule, wherein the modification is conducted at a temperature that is suitable for modification.

2. The method of claim 1, wherein the modification is an enzymatic modification with at least a first enzyme, and the apparatus is maintained for a first period of time at a first temperature at which the first enzyme exhibits at least 10% of its maximal activity.

3. The method of claim 2, further comprising:

c) modifying the immobilized biomolecule with a second enzyme, wherein the apparatus is maintained for a second period of time at a second temperature at which the second enzyme exhibits at least 10% of its maximal activity.

4. The method of claim 1, further comprising eluting the modified biomolecule from the column.

5. The method of claim 1, wherein the biomolecule comprises a polypeptide, and the modification is selected from the group consisting of phosphorylation, dephosphorylation, nitrosylation, acetylation, deglycosylation, glycosylation, acylation, methylation, ADP riboxlation, ubiquitination, lipidation, carboxylation, hydroxylation, and nucleotidylation.

6. The method of claim 1, wherein the biomolecule comprises a polypeptide, and the modification is labeling with a detectable label.

7. The method of claim 1, wherein the immobilized biomolecule comprises a polynucleotide, and the modification comprises hybridization to a second biomolecule

comprising a polynucleotide comprising a nucleotide sequence that is substantially complementary to at least a portion of the immobilized polynucleotide.

8. The method of claim 7, wherein the immobilized biomolecule is a polynucleotide, and the modification comprises synthesizing a polynucleotide comprising a nucleotide sequence that is complementary to a nucleotide sequence in the immobilized polynucleotide.

9. The method of claim 1, wherein the immobilized biomolecule comprises a polynucleotide, and the modification is an enzymatic modification selected from the group consisting of synthesis of a polynucleotide complementary to the immobilized polynucleotide, addition of a nucleotide to the 5' end of the immobilized polynucleotide, addition of a nucleotide to the 3' end of the immobilized polynucleotide, ligation of a single-stranded polynucleotide to the immobilized polynucleotide, ligation of a double-stranded polynucleotide to the immobilized polynucleotide, cleavage of the immobilized polynucleotide at a restriction endonuclease recognition site, removal of a nucleotide from the immobilized polynucleotide, synthesis of a polypeptide using the immobilized polynucleotide as a template, and methylation of a base of a nucleotide of the immobilized polynucleotide.

10. The method of claim 1, wherein the immobilized biomolecule comprises a polynucleotide, and the modification is a non-enzymatic modification.

11. The method of claim 1, wherein the immobilized biomolecule comprises a polynucleotide, and the modification comprises binding a polypeptide to the immobilized polynucleotide.

12. The method of claim 1, wherein the immobilized biomolecule comprises a first polypeptide, and the modification comprises binding a second polypeptide to the immobilized polypeptide.

13. The method of claim 1, wherein the immobilized biomolecule comprises a double-stranded polynucleotide, and the modification comprises contacting the immobilized

polynucleotide with a double-stranded polynucleotide of from about 6 to about 20 nucleotides in length, in the presence of a DNA ligase, at a temperature of about 16°C.

14. A method of synthesizing a nucleic acid molecule, comprising:

a) immobilizing a biomolecule bound to a magnetic particle on a magnetic separation apparatus by applying a magnetic field to a magnetizable matrix in the column, wherein the immobilized biomolecule comprises a polynucleotide and wherein the magnetic particle contains bound thereto an oligonucleotide that is complementary to a portion of the immobilized biomolecule and that serves as a primer for synthesis of a nucleic acid;

b) contacting the immobilized polynucleotide with an enzyme that can synthesize a nucleic acid molecule, in the presence of deoxynucleotides, wherein the apparatus is maintained for a period of time at a temperature at which the enzyme exhibits at least 10% of its maximal activity; and

c) synthesizing a nucleic acid molecule, using the immobilized polynucleotide as a template.

15. The method of claim 14, wherein at least one deoxynucleotide comprises a detectable label, and wherein the synthesized nucleic acid molecule comprises the at least one detectably labeled deoxynucleotide.

16. The method of claim 14, wherein the immobilized polynucleotide is an mRNA molecule, wherein the enzyme is a reverse transcriptase, wherein step (c) is conducted at a temperature of from about 32°C to about 42°C, and wherein the synthesized nucleic acid molecule is a cDNA molecule.

17. The method of claim 16, further comprising:

d) contacting the cDNA molecule with RNaseH at a temperature of about 37°C; and

e) eluting the cDNA molecule.

18. A method of synthesizing a nucleic acid molecule, comprising:

a) immobilizing a biomolecule bound to a magnetic particle on a magnetic separation apparatus by applying a magnetic field to a magnetizable matrix in the column, wherein the immobilized biomolecule comprises a polynucleotide;
b) contacting the immobilized polynucleotide with a first oligonucleotide primer and an enzyme that can synthesize a nucleic acid molecule, in the presence of deoxynucleotides, wherein the apparatus is maintained for a period of time at which the enzyme exhibits at least 10% of its maximal activity; and
c) synthesizing a nucleic acid molecule, using the immobilized polynucleotide as a template.

19. The method of claim 18, wherein step (b) is conducted at a temperature of about 55°C, and wherein step (c) is conducted at a temperature of from about 60°C to about 72°C.

20. The method of claim 18, further comprising:
d) heating the column to a temperature of from about 90°C to about 96°C;
e) contacting the synthesized nucleic acid molecule with a second oligonucleotide primer that hybridizes to a region in the synthesized nucleic acid molecule;
f) bringing the column to about 55°C for a period of time sufficient to allow hybridization of the second primer to the synthesized nucleic acid molecule; and
g) bringing the column to a temperature of from about 60°C to about 72°C.

21. The method of claim 18, comprising repeating steps (d), (f), and (g) from 2 to about 30 times.

22. A method of synthesizing a nucleic acid molecule, comprising:
a) immobilizing a biomolecule bound to a magnetic particle on a magnetic separation apparatus by applying a magnetic field to a magnetizable matrix in the column, wherein the immobilized biomolecule comprises a polynucleotide comprising a poly(A) tract and the magnetic particle is bound to an oligo-dT molecule of from about 6 nucleotides to about 30 nucleotides;
b) contacting the immobilized polynucleotide with an enzyme that can synthesize a nucleic acid molecule, in the presence of deoxynucleotides, wherein the

apparatus is maintained for a period of time at a temperature at which the enzyme exhibits at least 10% of its maximal activity; and

c) synthesizing a nucleic acid molecule, using the immobilized polynucleotide as a template.

23. The method of claim 22, wherein the immobilized polynucleotide is an mRNA molecule, and the synthesized nucleic acid molecule is a cDNA molecule.

24. The method of claim 23, further comprising contacting the immobilized mRNA and the synthesized cDNA molecule with RNAseH.

25. The method of claim 22, wherein at least one of the deoxynucleotides comprises a detectable label, wherein the detectably labeled deoxynucleotide is incorporated into the synthesized nucleic acid molecule.

26. A system for immobilizing and modifying biomolecules, comprising:

at least one separation chamber;

a wettable, flow through heat conducting matrix contained in each said separation chamber; and

a controllable heat source thermally coupled to each said separation chamber.

27. The system of claim 26, further comprising a controllable cooling source coupled to each said separation chamber.

28. The system of claim 27, wherein each said controllable heat source also functions as said controllable cooling source, respectively.

29. The system of claim 26, further comprising a controller coupling each said controllable heat source with a power source, wherein said controller functions to control an amount of power delivered to each said controllable heat source to control a temperature thereof.

30. The system of claim 29, further comprising a feedback sensor associated with each said controllable heat source to provide feedback to said controller regarding a temperature of said respective controllable heat source.

31. The system of claim 30, wherein each said feedback sensor comprises a thermocouple.

32. The system of claim 26, wherein said wettable, flow through heat conducting matrix is internally magnetizable.

33. The system of claim 26, wherein said controllable heat source comprises at least one heating film.

34. The system of claim 26, wherein said controllable heat source comprises at least one power resistance type heating element

35. The system of claim 26, wherein said controllable heat source comprises at least one Peltier element.

36. The system of claim 26, wherein said controllable heat source comprises a pneumatic heating system.

37. The system of claim 26, wherein said controllable heat source comprises a hydraulic heating system.

38. The system of claim 26, wherein said controllable heat source comprises at least one radiant heating element.

39. The system of claim 38, wherein each said radiant heating element comprises an infrared light emitting diode.

40. The system of claim 26, wherein said controllable heat source comprises at least one inductive heating element.

41. The system of claim 26, wherein each said inductive heating element comprises a spool of wound wire.

42. A method for modifying a biomolecule, comprising:

a) immobilizing a biomolecule bound to a magnetic particle on a system according to claim 26 by applying a magnetic field to a magnetizable matrix in the separation chamber; and

b) modifying the immobilized biomolecule, wherein the modification is conducted at a temperature that is suitable for modification.

43. A separation unit for immobilizing and modifying biomolecules, comprising:

a magnetic yoke having at least one notch formed therein:

a pair of magnets placed within each of said at least one notch to form a gap therebetween, said gap being adapted to receive a separation chamber therein; and

a controllable heat source thermally coupled to each said pair of magnets.

44. The unit of claim 43, further comprising an insulation layer separating said magnets and said controllable heat source.

45. The unit of claim 43, wherein each said controllable heat source also functions as a controllable cooling source.

46. The unit of claim 43, further comprising a heat conducting element thermally connecting each said controllable heating source with said respective pair of magnets.

47. The unit of claim 46, wherein each said heat conducting element is configured to contact a separation chamber for conducting heat thereto.

48. The unit of claim 43, wherein at least one of said controllable heat sources comprises a heating film.

49. The unit of claim 43, wherein at least one of said controllable heat sources comprises a Peltier type heating source.

50. The unit of claim 43, wherein at least one of said controllable heat sources comprises a power resistance type heating source.

51. The unit of claim 43, wherein at least one of said controllable heat sources comprises a liquid driven element adapted to transfer heat from a liquid circulated therethrough.

52. The unit of claim 43, wherein at least one of said controllable heat sources comprises a radiant heating element.

53. The system of claim 52, wherein each said radiant heating element comprises an infrared light emitting diode.

54. The system of claim 43, wherein at least one of said controllable heat sources comprises an inductive heating element.

55. The system of claim 54, wherein each said inductive heating element comprises a spool of wound wire.

56. The unit of claim 43, further comprising a controller coupling each said controllable heat source with a power source, wherein said controller functions to control an amount of power delivered to each said controllable heat source to control a temperature thereof.

57. The unit of claim 54, further comprising a feedback sensor associated with each said controllable heat source to provide feedback to said controller regarding a temperature of said respective controllable heat source.

58. The unit of claim 57, wherein each said feedback sensor comprises a thermocouple.

59. A separation unit for immobilizing and modifying biomolecules, comprising:

a magnetic yoke having at least one notch formed therein:

a pair of magnets placed within each of said at least one notch to form a gap therebetween, said gap being adapted to receive a separation chamber therein; and

a controllable cooling source thermally coupled to each said pair of magnets.

60. An external temperature regulating unit adapted to interface with an HGMS separation unit, said regulating unit comprising:

a base portion;

finger elements extending from said base portion and adapted to fit within slots in the HGMS separation unit which hold separation columns; and

a controllable heating element at an end of each said finger element, adapted to apply a controlled amount of heat to the separation column in the gap, respectively.

61. The regulating unit of claim 60, wherein said base portion comprises a heat conductor made of a heat conducting material.

62. The regulating unit of claim 61, wherein said finger elements are formed of the same heat conducting material as said heat conductor.

63. The regulating unit of claim 60, wherein each said finger element has a width which is substantially the same as the width of the gap into which it is to be inserted, so that the finger substantially fills the remainder of the gap that is left after the column is inserted in the gap.

64. The regulating unit of claim 60, wherein each said finger element has a length sufficient to allow contact between an end of said finger element and the column while maintaining said base portion in close approximation with the separation unit.

65. The regulating unit of claim 60, wherein each said finger element comprises an end having a concave surface adapted to abut and closely interface with a portion of the circumference of the respective column in the gap.